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An antigenic determinant (epitope) unique to a single clone of cells and located in the variable region of the immunoglobulin product of that clone or to the T-cell receptor. The idiotope forms part of the antigen binding site. Any single immunoglobulin may have more than one idiotope. Idiotopes are also associated with the antigen binding sites of T-cell receptors. They are the epitopes to which an anti-idiotypic antibody or T-cell binds.

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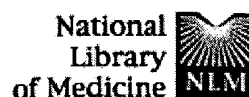
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## **IgA:IgM and IgA:IgA hybrid hybridomas secrete heteropolymeric immunoglobulins that are polyvalent and bispecific.**

**Urnovitz HB, Chang Y, Scott M, Fleischman J, Lynch RG.**

Department of Pathology, University of Iowa College of Medicine, Iowa City 52242.

Polyvalent bispecific antibodies were secreted by hybrid hybridoma cells when both parental clones expressed a naturally polymerizing immunoglobulin. Hybrid hybridomas made from IgA lambda 2 anti-trinitrophenyl (TNP) and IgA kappa anti-phosphocholine (PC) parental cells secreted polymeric IgA antibodies that bound both TNP and PC. Some of the TNP binding was dissociated from the PC binding under conditions of mild reduction and alkylation suggesting that the bispecific polymeric IgA contained disulfide-linked parental monomers as well as bispecific hybrid monomers. Hybrid hybridomas constructed from IgA lambda 2 anti-TNP and IgM kappa anti-ox erythrocyte parental cells secreted bispecific, polymeric immunoglobulin that contained mu-, alpha-, kappa-, and lambda 2-chains. The mu and kappa-chains dissociated from the alpha- and lambda 2-chains under conditions of mild reduction and alkylation, indicating that both parental monomers had been incorporated into the same polymeric immunoglobulin to form a heteropolymeric antibody molecule. Heterologous pairing of alpha and mu heavy chains in monomers was not detected. Hybrid hybridomas constructed from IgA lambda 2 and IgG3 lambda 2 or IgA lambda 2 and IgG1 kappa parents co-secreted both parental immunoglobulins, but the antibodies secreted by these clones did not form heteropolymers or exhibit heterologous heavy chain pairing. These findings establish that polyvalent, bispecific, polymeric immunoglobulin molecules can be produced by hybrid hybridomas when both parents express a naturally polymerizing class of heavy chain but not when only one parent does. Hybrid hybridomas that produce heteropolymeric immunoglobulins are sources of high avidity bispecific antibodies that may find a number of basic and practical applications. The hybridoma cells that produce these antibodies may provide useful tools for investigating the in situ determinants of immunoglobulin chain association and the regulation of antibody assembly and secretion.

The term "active enzyme" refers to an enzyme which is functional (*i.e.*, capable of carrying out the enzymatic function).

Immunoglobulin molecules consist of heavy (H) and light (L) chains, which comprise highly specific variable regions at their amino termini. The variable (V) regions of the H ( $V_H$ ) and L ( $V_L$ ) chains combine to form the unique antigen recognition or antigen combining site of the immunoglobulin (Ig) protein. The variable regions of an Ig molecule contain determinants (*i.e.*, molecular shapes) that can be recognized as antigens or idiotypes.

The term "idiotype" refers to the set of antigenic or epitopic determinants (*i.e.*, idiotopes) of an immunoglobulin V domain (*i.e.*, the antigen combining site formed by the association of the complementarity determining regions or  $V_H$  and  $V_L$  regions).

The term "idiotope" refers to a single idiotypic epitope located along a portion of the V region of an immunoglobulin molecule.

The term "anti-idiotypic antibody" or grammatical equivalents refers to an antibody directed against a set of idiotopes on the V region of an Ig protein.

A "multivalent vaccine" when used in reference to a vaccine comprising an idiotypic protein or fragment thereof (*e.g.*, immunoglobulin molecules or variable regions thereof, T cell receptor proteins or variable regions thereof) refers to a vaccine which contains at least two idiotypic proteins which differ by at least one idiotope. For example, a vaccine which contains two or more immunoglobulin molecules derived from a B-cell lymphoma where the immunoglobulin molecules differ from one another by at least one idiotope (*e.g.*, these immunoglobulins are somatic variants of one another) is a multivalent vaccine.

As used herein "recombinant variable regions of immunoglobulin molecules" refers to variable regions of Ig molecules which are produced by molecular biological means. As shown herein, the variable domain of the heavy and light chains may be molecularly cloned from lymphoma cells and expressed in a host cell (*e.g.*, by insertion into an expression vector followed by transfer of the expression vector into a host cell); variable domains expressed in this manner are recombinant variable regions